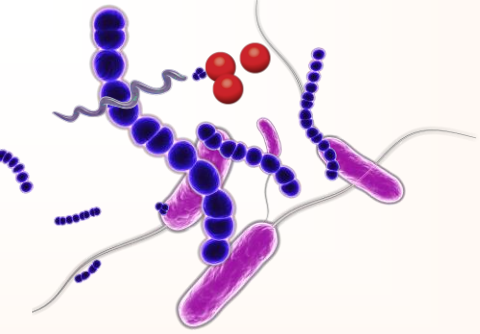


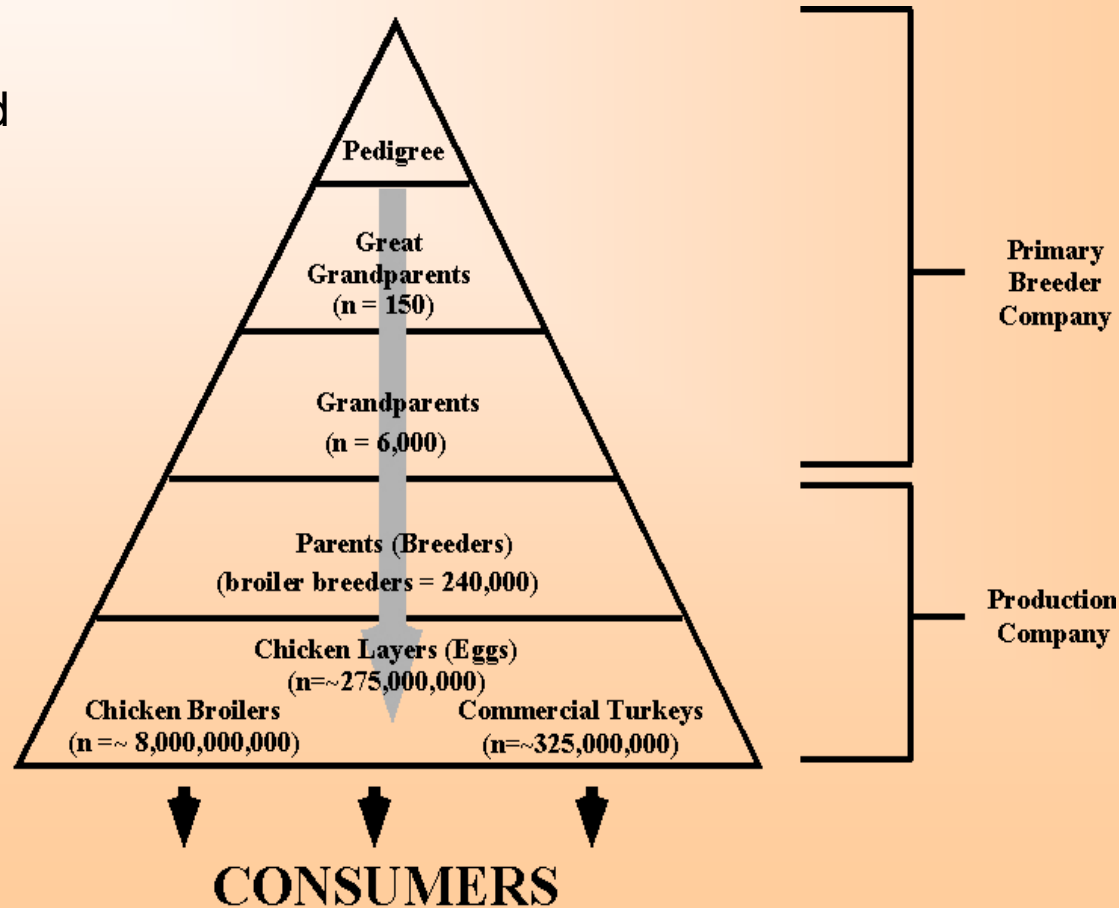
# ***The Chicken Intestinal Microbiome as a Target for Improving Productivity***

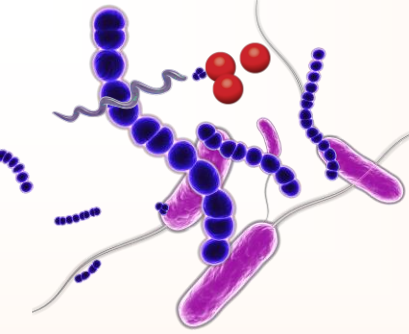
**Margie D. Lee D.V.M. Ph.D.  
Department of Population Health  
Poultry Diagnostic and Research Center  
College of Veterinary Medicine  
The University of Georgia**



# Modern Commercial Poultry Production

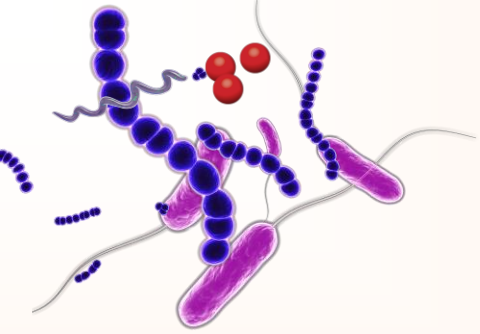
Genetics of rapid growth and improved feed conversion





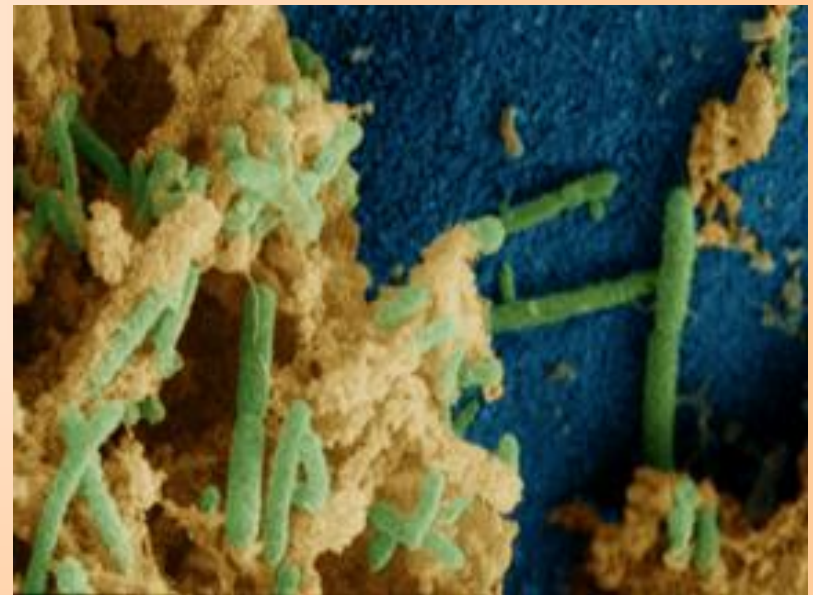
# Alternatives to Antibiotics to modify microbiome

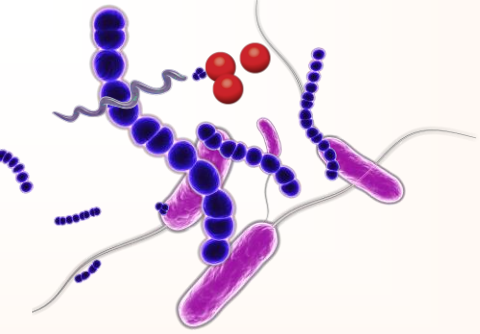
- Poultry scientist - science of nutrition
  - Improved feed efficiency
  - Improved growth rates
  - More uniformity in size within flock
  - Reduce shedding of pathogens
- Veterinarians – science of disease
  - Prevent intestinal diseases
  - Reduce inflammation
  - Prevent colonization with foodborne pathogens
  - Reduce shedding of pathogens



# Intestinal Microbiome

- Bacteria, fungi, viruses, protozoa, helminths
- Bacteria  $10^{11}$  cells/gram
- Bacteria primarily associated with mucus and macromolecular food matrix (fiber)
- Composition varies
  - in different portions of GI
  - in different animals

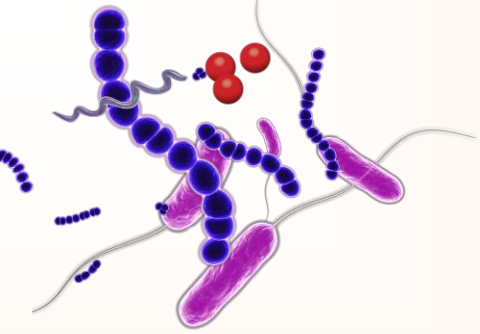




# Intestinal Microbiome

(lessons from germ-free animals)

- Stimulates intestinal maturation
- Stimulation of immunity
- Degrades mucus
- Inhibition of pathogens
- Growth promotion
  - Degradation of nondigestible foodstuffs
  - Volatile fatty acid production
  - Vitamin production



# Models of Host/Microbe Symbiosis



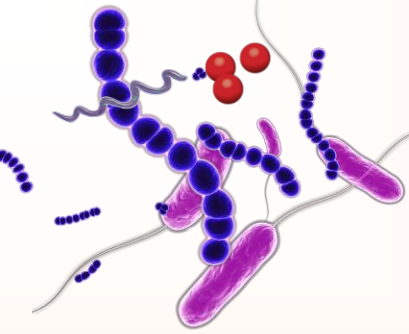
Ruminant physiology –  
digestion of cellulose



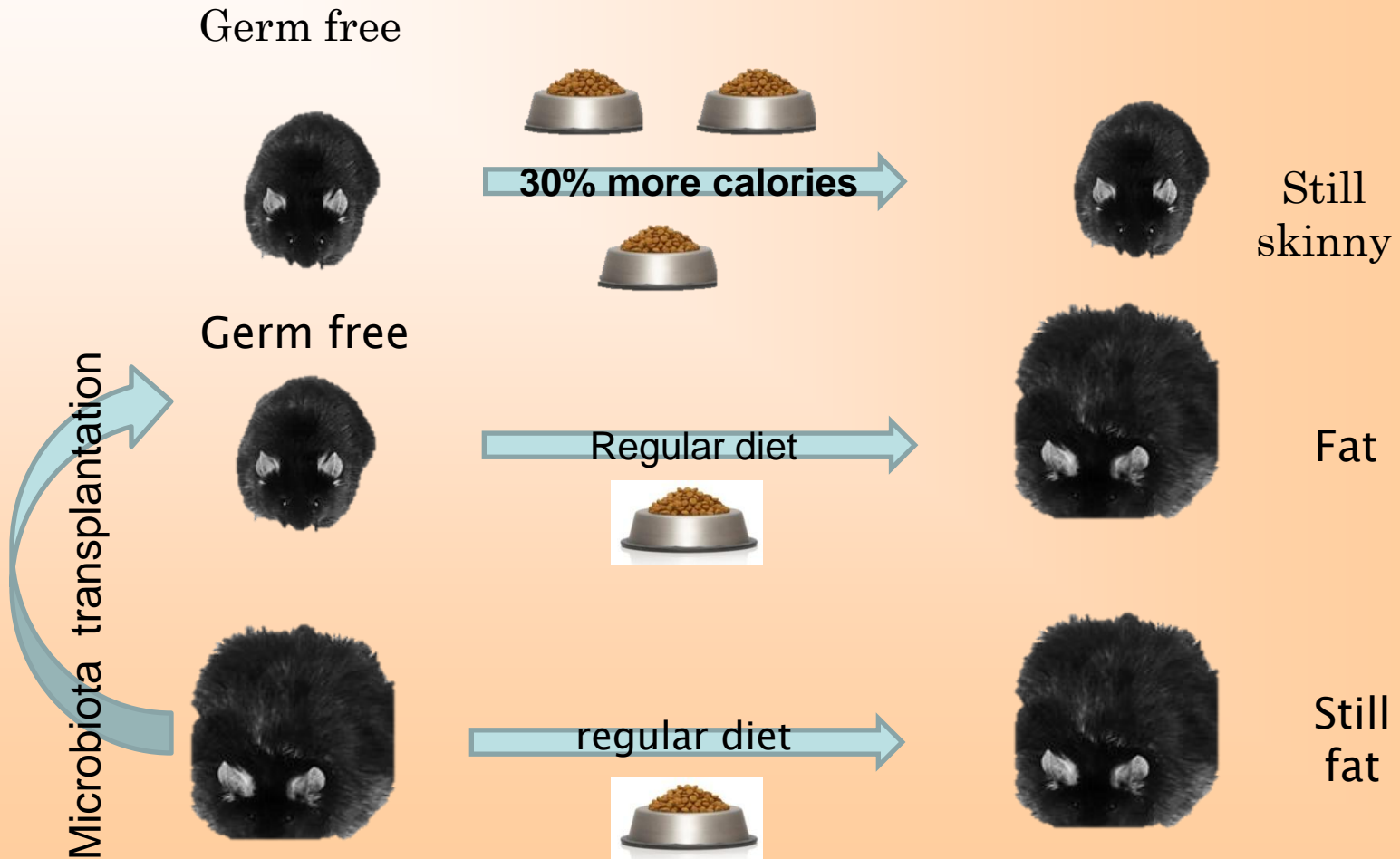
Termite physiology –  
digestion of wood



Marine animal physiology  
- Bioluminescence

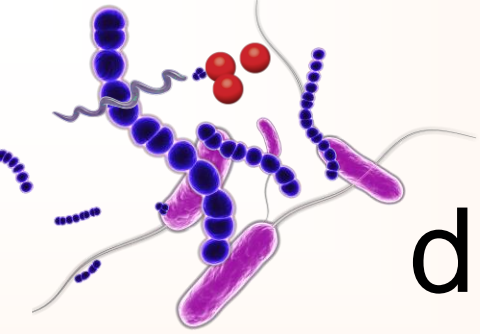


# Microbiome may regulate energy storage

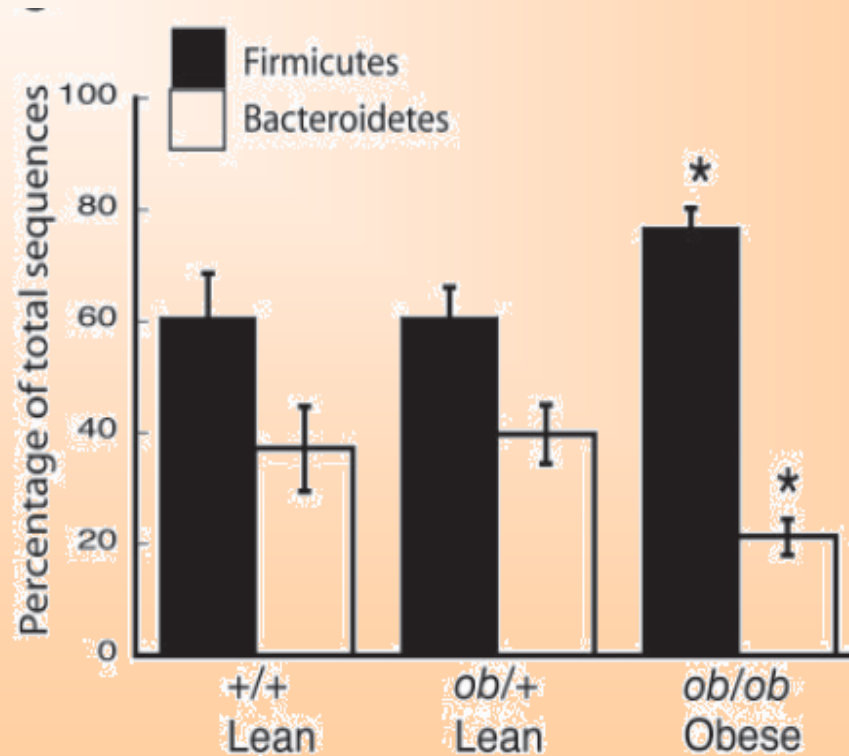


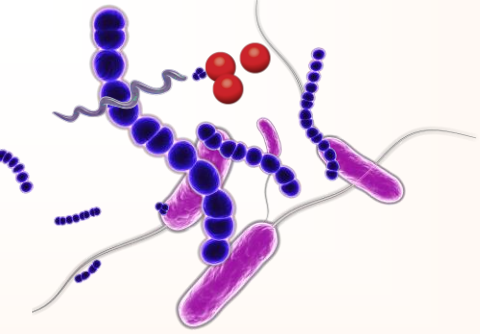
Conventional

Backhed et al, 2004



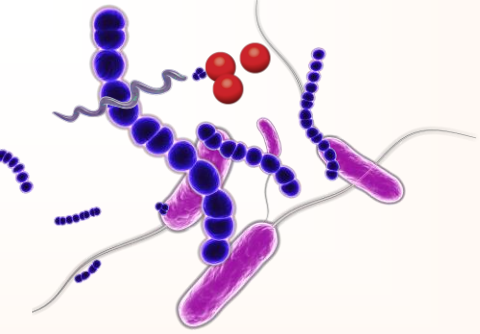
# Cecal microbiome is different in genetically lean and fat mice



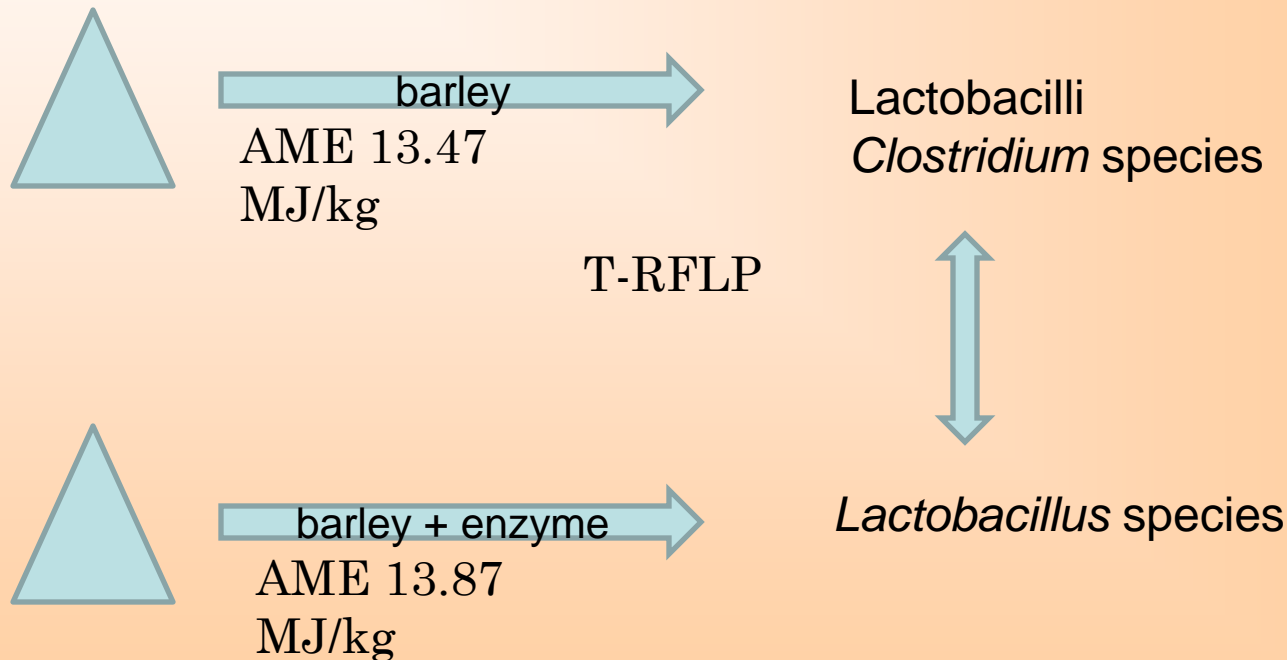


# Intestinal microbiome ferments available polysaccharides

- Fermentation -> acetate, propionate and butyrate (short chain fatty acids)
- Energy for intestinal cells
- Substrate for lipid synthesis by epithelial cells



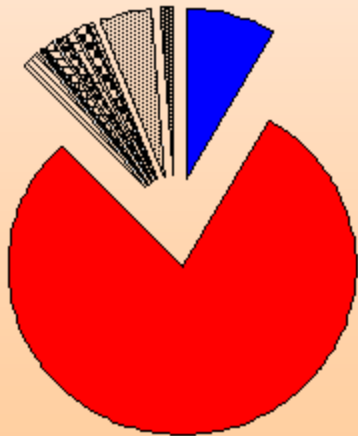
# Chicken microbiome is linked to energy metabolism



Torok et al., 2008

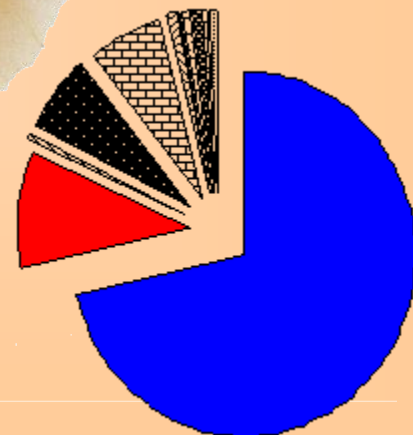
# Microbial Composition of the Broiler Intestine (16S based)

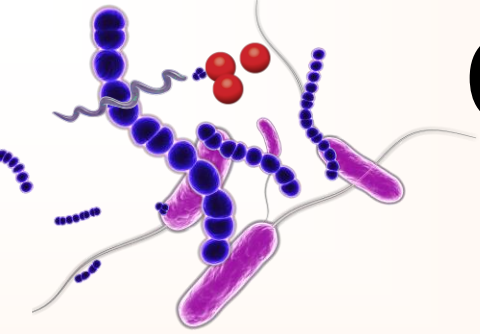
**Cecum**



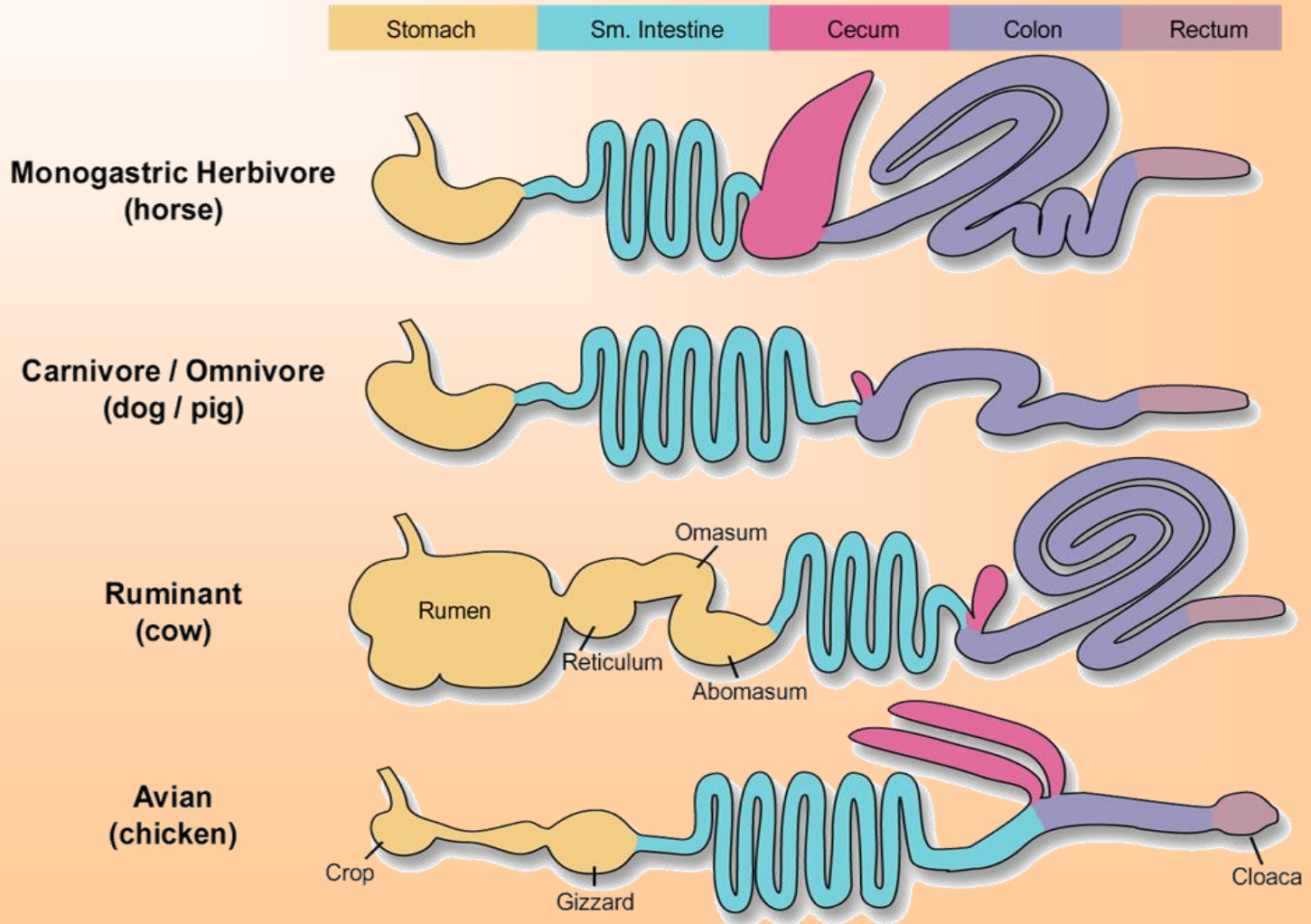
- Lactobacillaceae (8.5%)
- Clostridia (82%)
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- Streptococcaceae (0.7%)
- Enterococcaceae (1.0%)
- Actinobacteria (0%)
- Alpha (0.8%)
- Beta (0.7%)
- Gamma (1.3%)
- Flavobacteriaceae (0.2%)
- Bacteroidaceae (5.1%)
- Unknown bacteria (1.1%)

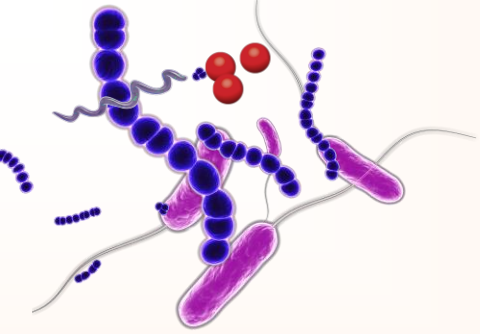
**Ileum**





# Comparative anatomy of intestinal tract





# Models of Host/Microbe Symbiosis



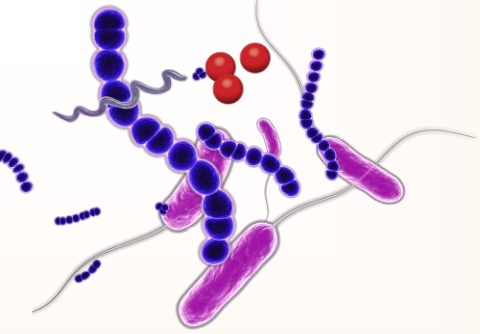
Ruminant physiology –  
digestion of cellulose



Termite physiology –  
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Marine animal physiology  
- Bioluminescence

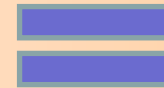


# Bacterial Symbiosis - bioluminescence

- Light-Organ Symbiosis of *Vibrio fischeri* and the Hawaiian squid, *Euprymna scolopes*



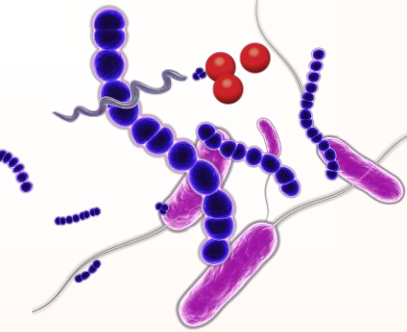
Hawaiian squid



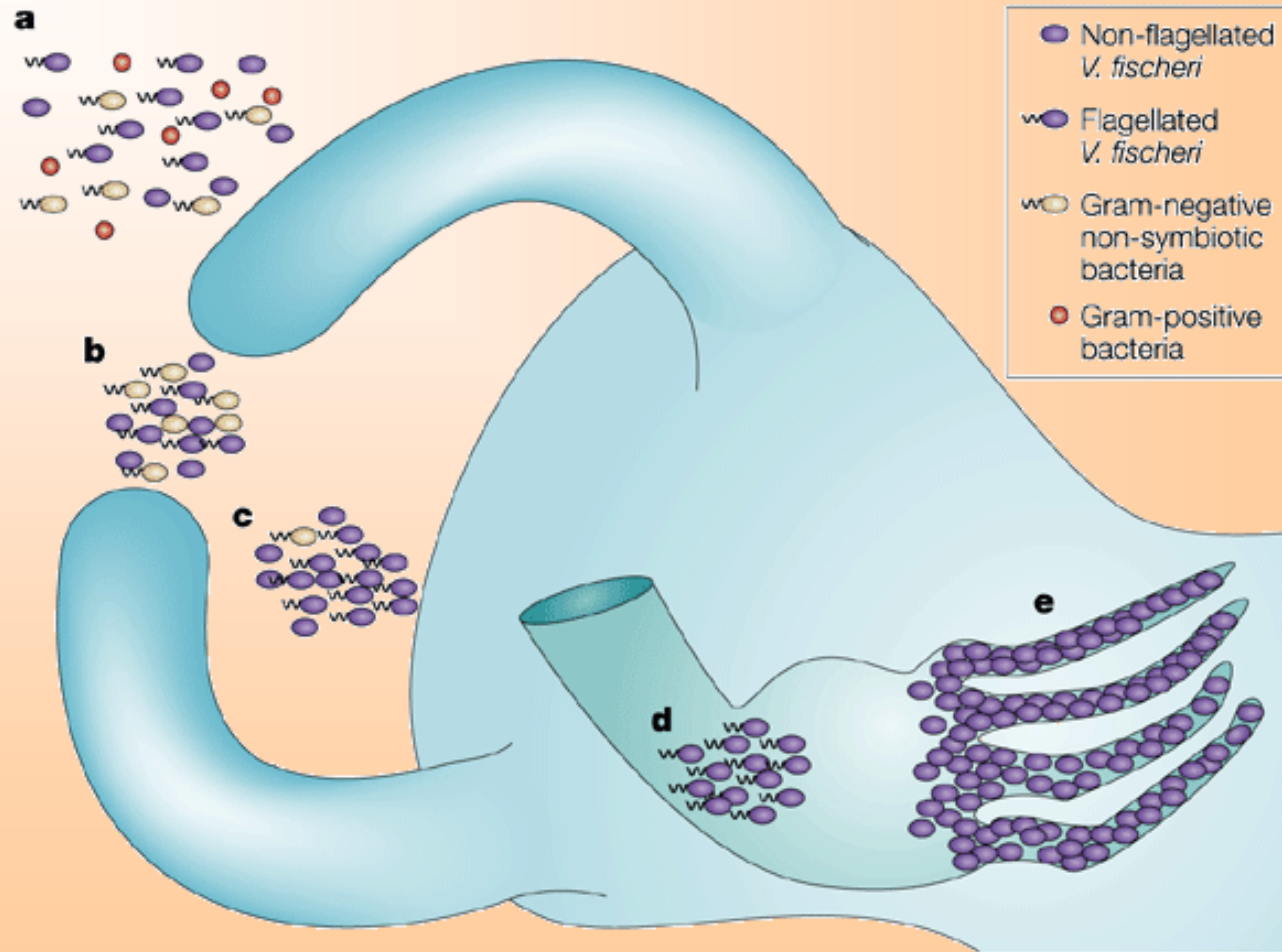
*Vibrio fischeri*  
colonizes the  
light organ

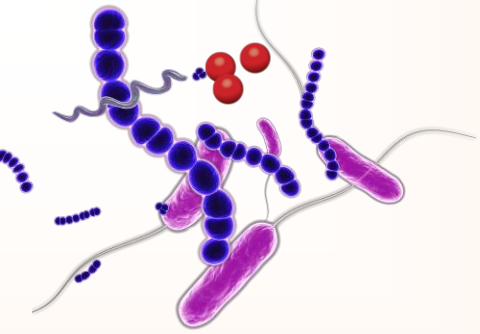


Luminescent  
squid



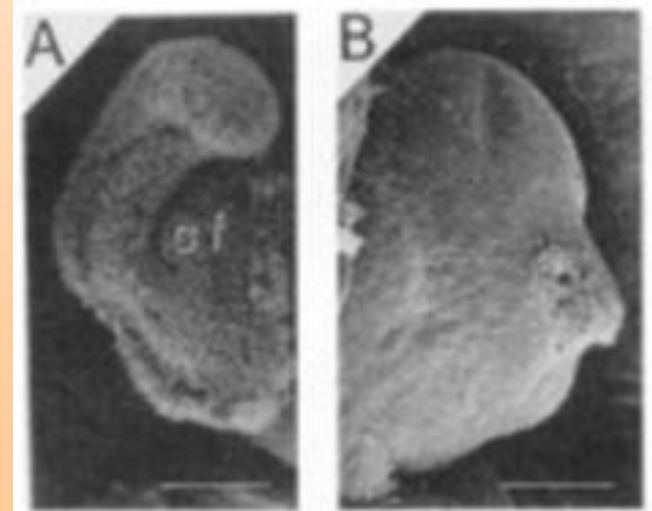
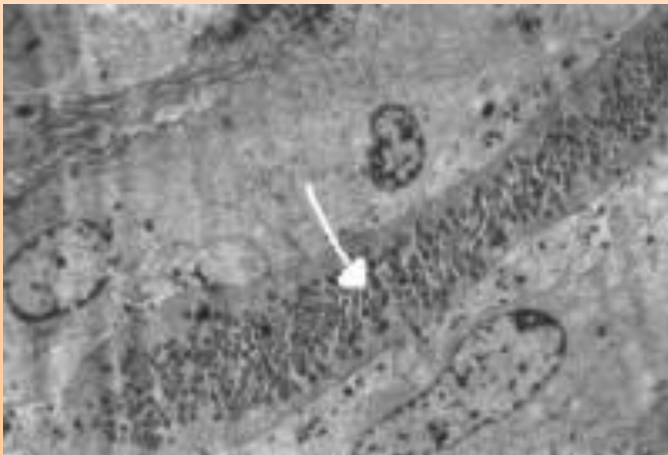
# Colonization of the light organ

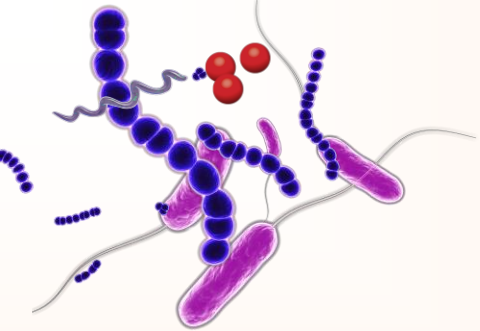




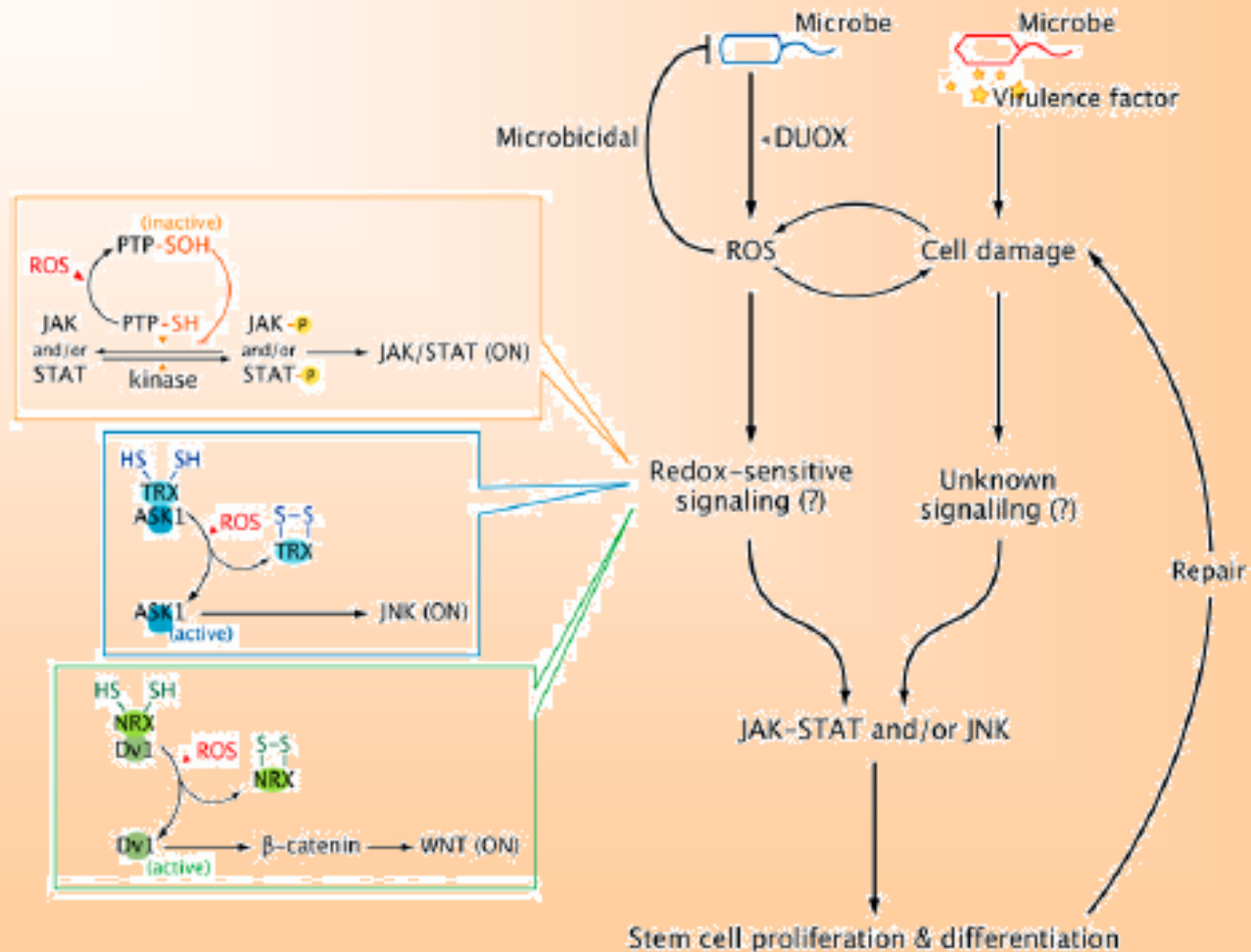
# Symbiosis or Disease?

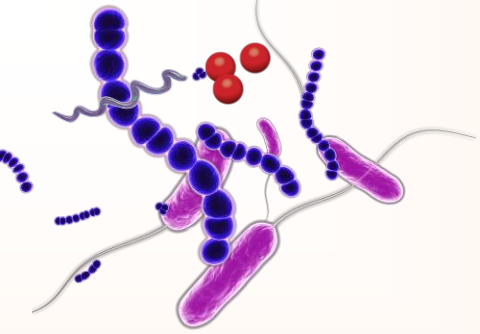
- *Vibrio* colonizes horn of light organ produces a toxin that causes inflammation
- Inflammation changes the anatomy of the light organ to better support its function





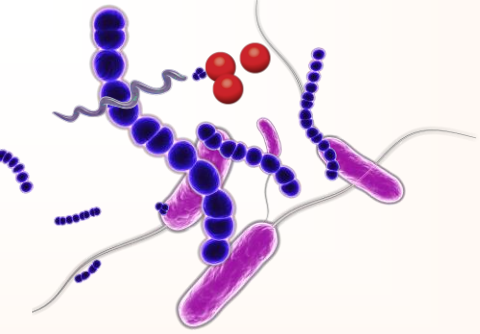
# Symbiosis or Disease?





# Intestinal Microbiome

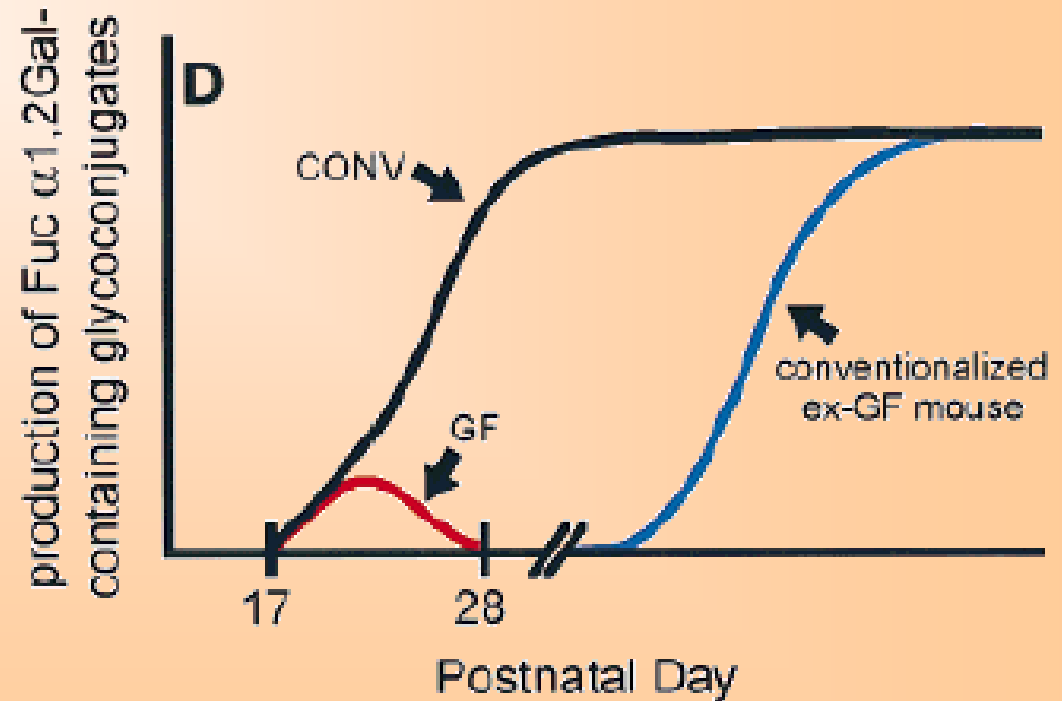
- Symbiosis – does it occur in the intestine?
- Commensalism – what role does this play in gut health?
- Pathogenicity – what controls pathogenic behavior in bacteria?

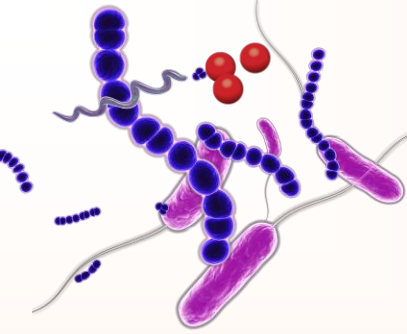


# Host–microbial symbiosis in the mammalian intestine: exploring an internal ecosystem

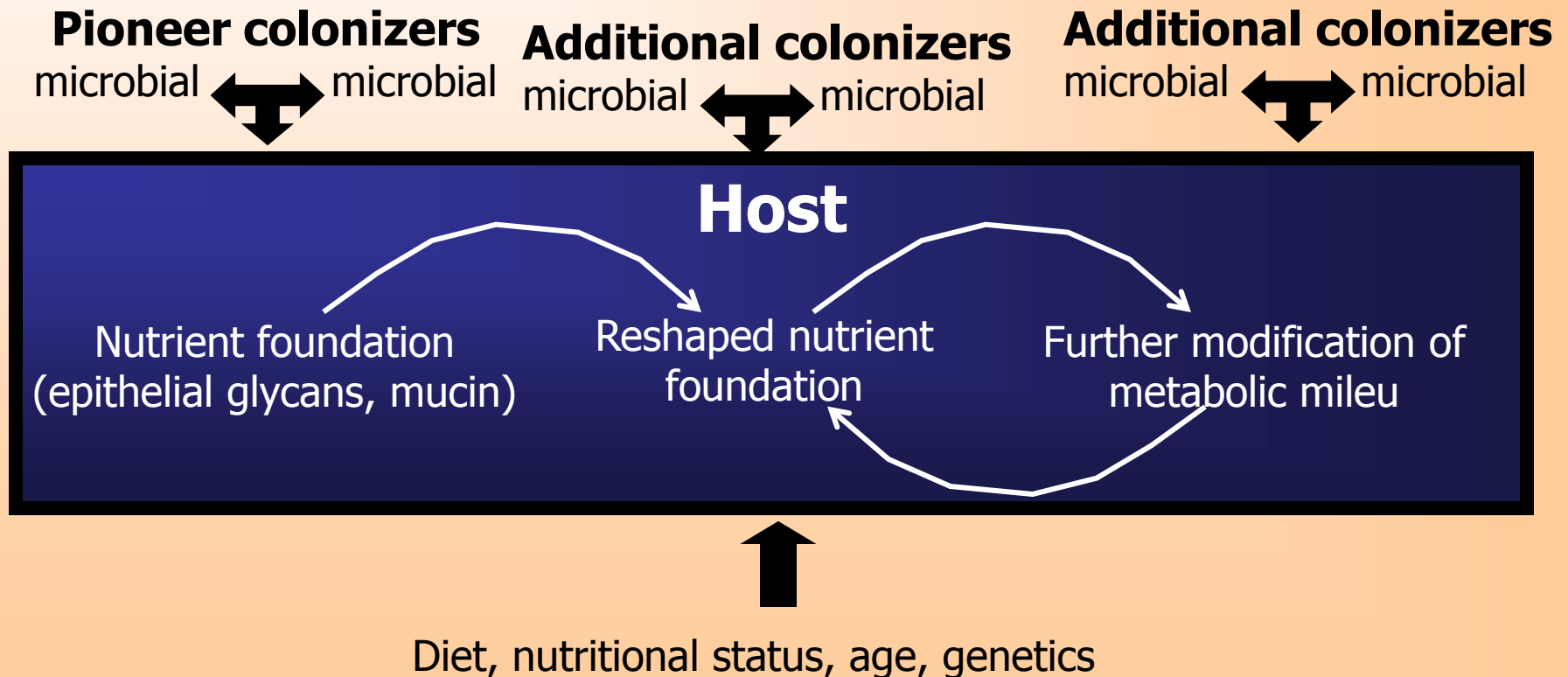
(Hooper et al. BioEssays 20:336–343, 1998)

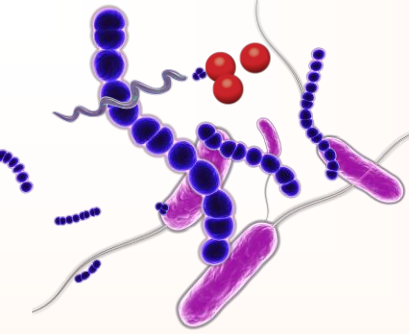
- Reconstitute gnotobiotic mice with conventional bacterial community
- Enhanced anatomical and functional development of the intestine
- Effects attributed to 1 member of the community = *Bacteroides thetaiotaomicron*



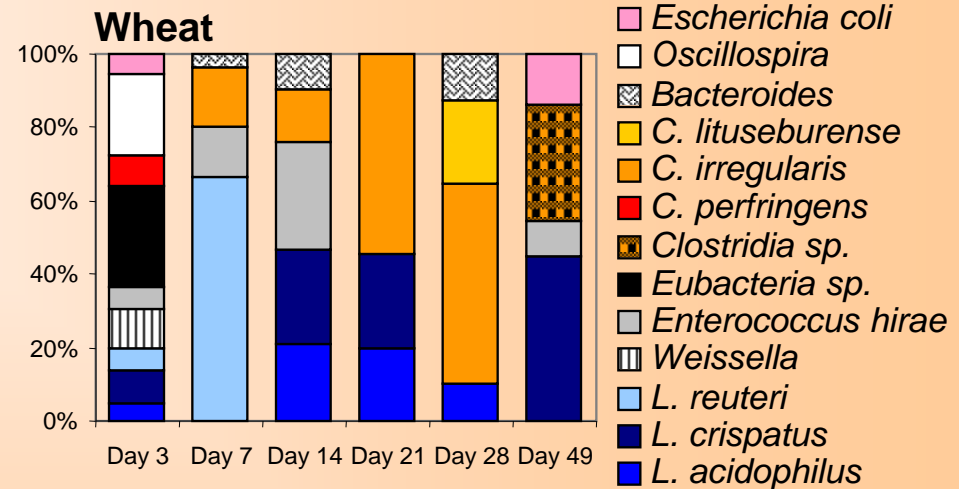
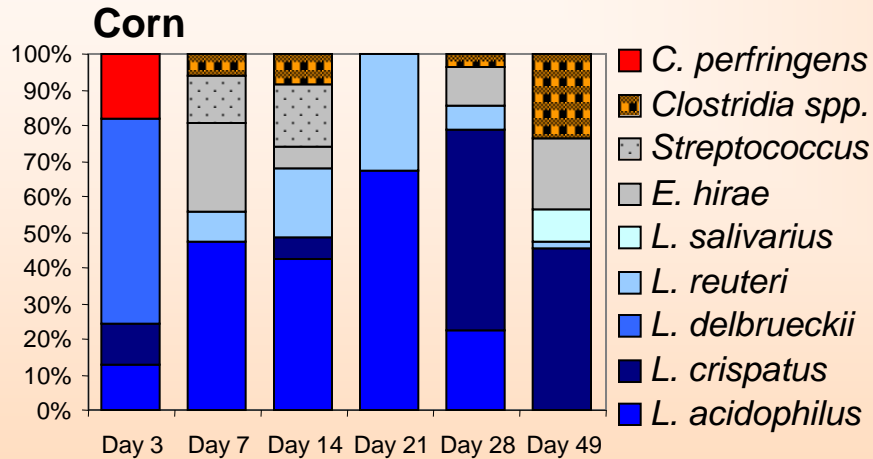


# Successional development of intestinal community



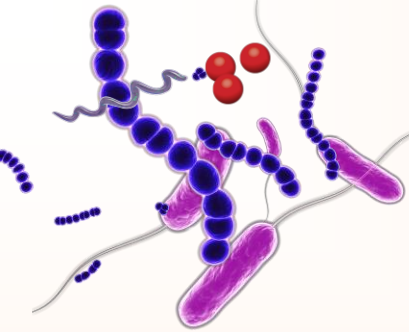


# Composition of broiler ileum (T-RFLP database analysis of 16S rRNA)

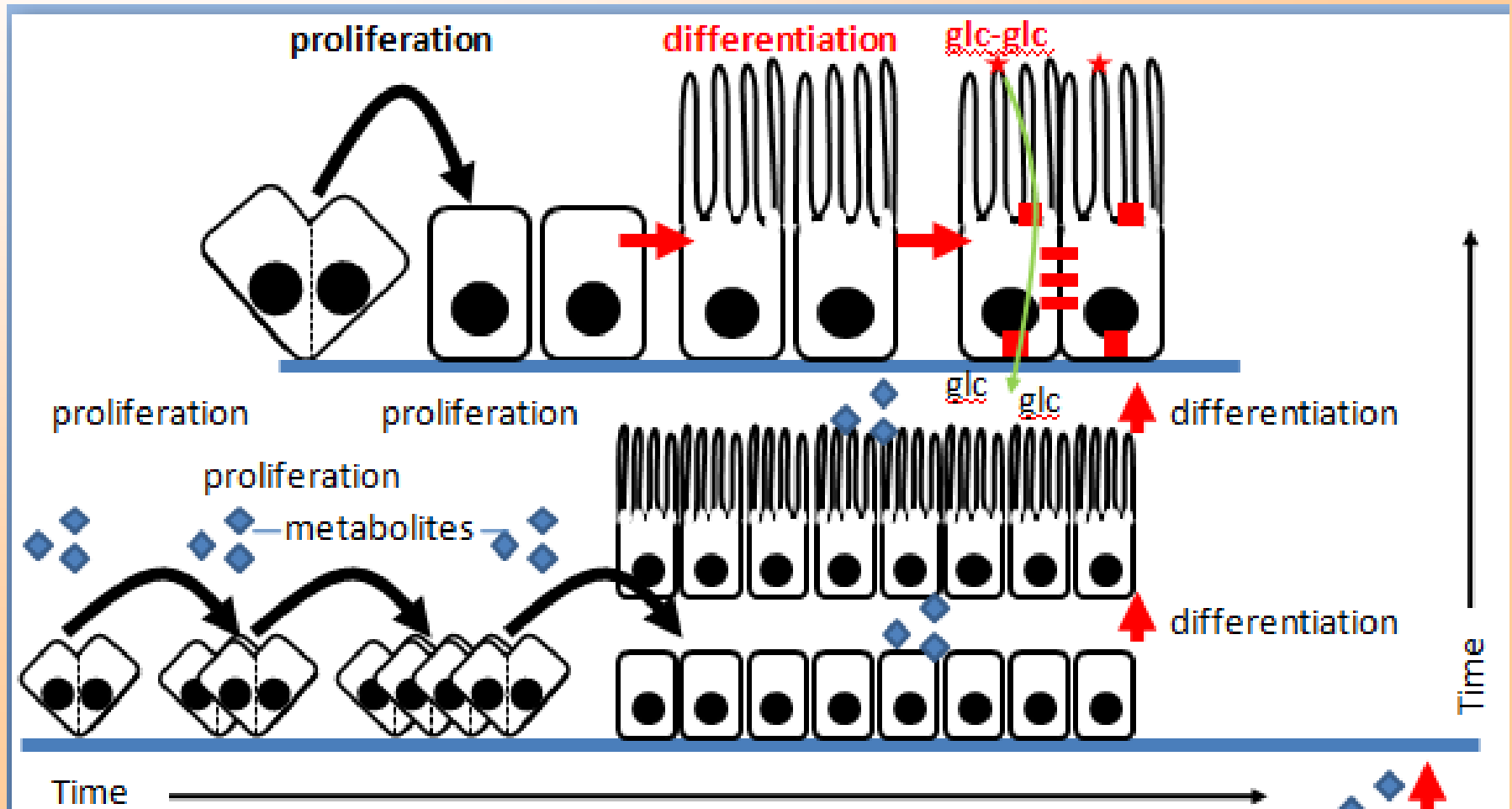


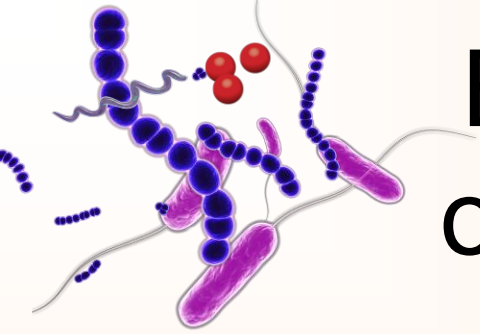
## Revealed Successional Periods

- age
- *Lactobacillus* species
- Clostridia

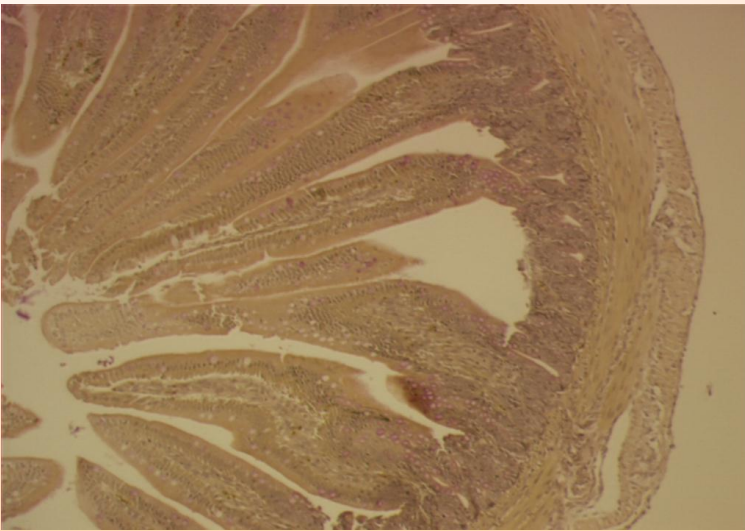


# Does the microbiome affect enterocyte differentiation?

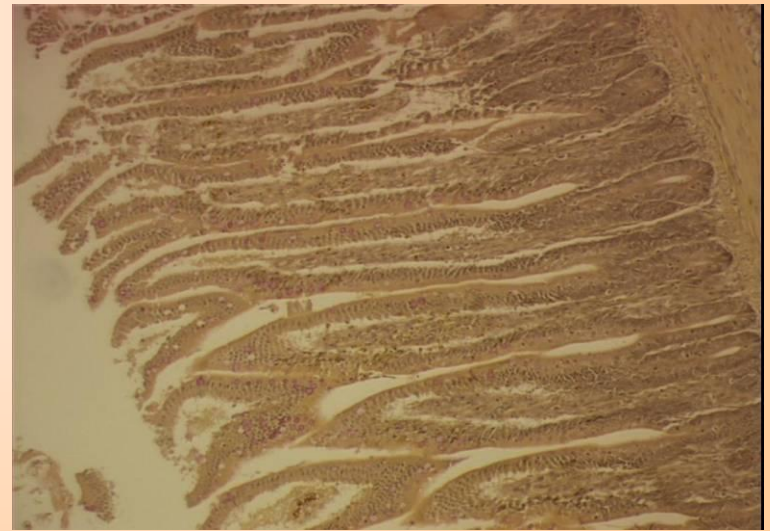




# Effect of pioneer colonizers on intestinal development of broiler chickens

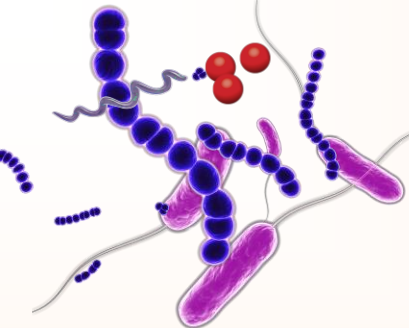


Control 2d



*Bacteroides* + *Clostridia* 2d

- Significantly different 1d, 7d, 16 days of age



# Effect of pioneer colonizers on intestinal development of broiler chickens

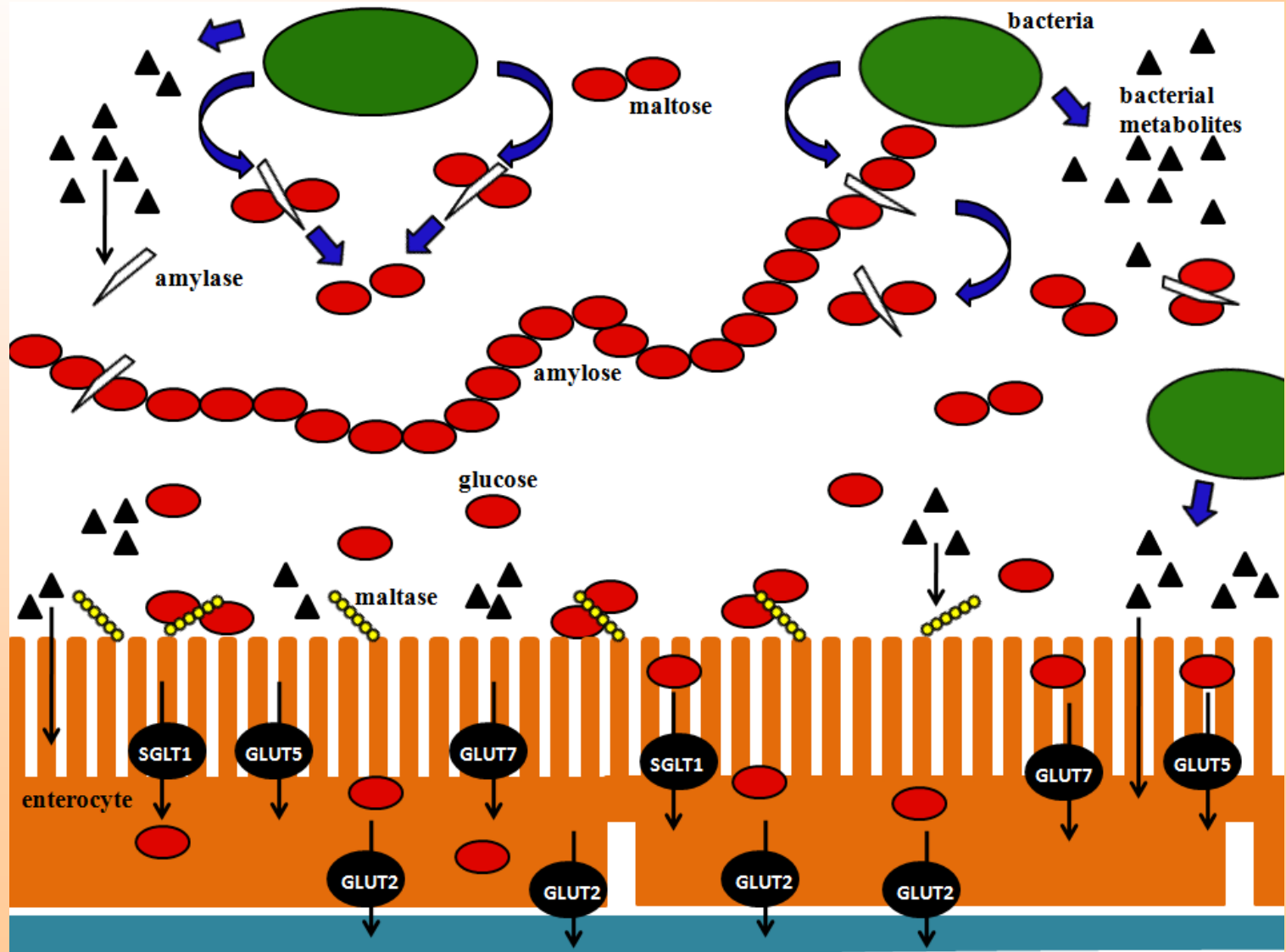
**Table 1.** Effects of probiotic bacteria orally administered to day of hatch chicks on villus height in the jejunum.

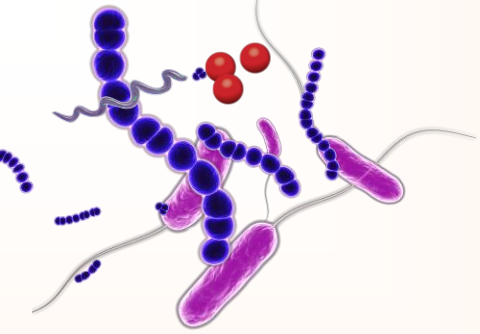
Treatments <sup>1</sup>	<i>Jejunum Villus Height</i> (µm)						
	0d	1d	2d	3d	7d	16d	42d
Control	149 <sup>b</sup>	205	246 <sup>ab</sup>	287 <sup>a</sup>	409 <sup>b</sup>	580 <sup>b</sup>	692
<i>Bacteroidaceae</i>	136 <sup>b</sup>	218	198 <sup>c</sup>	242 <sup>b</sup>	422 <sup>b</sup>	654 <sup>a</sup>	708
<i>Clostridiaceae</i>	124 <sup>b</sup>	187	226 <sup>bc</sup>	245 <sup>b</sup>	441 <sup>ab</sup>	645 <sup>a</sup>	732
both organisms	218 <sup>a</sup>	221	264 <sup>a</sup>	272 <sup>ab</sup>	467 <sup>a</sup>	668 <sup>a</sup>	694
Pooled Std Error of the Mear	12.3	9.4	15.2	10.9	13.6	23.2	23.2

Means represent 3 pens per treatment, 4 randomly selected chicks per pen, 3 villi per chick.

Means within a column and parameter without a common superscript differ significantly ( $P < 0.05$ ).

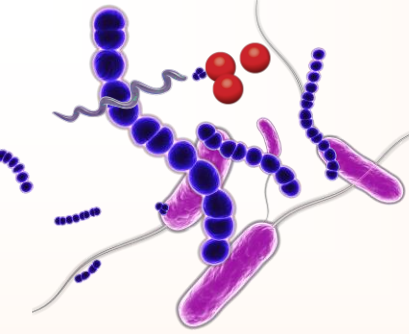
# Intestinal microbiome may affect nutrient acquisition





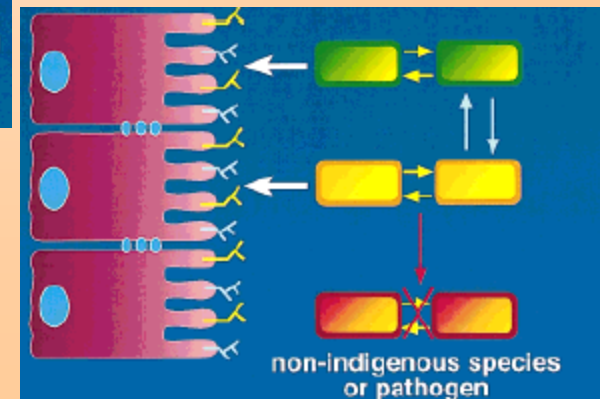
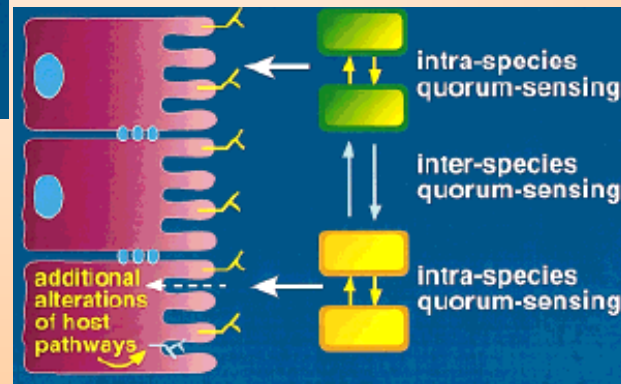
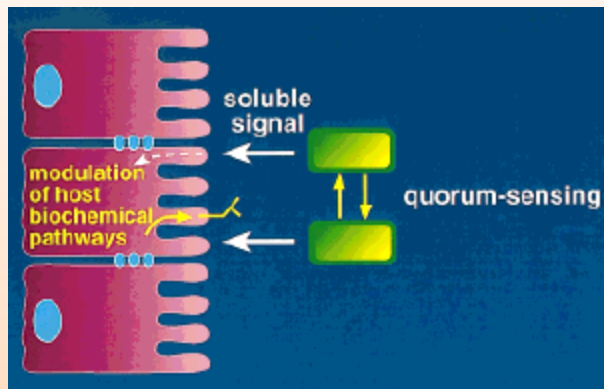
# Manipulation of Microbiome to Improve Performance

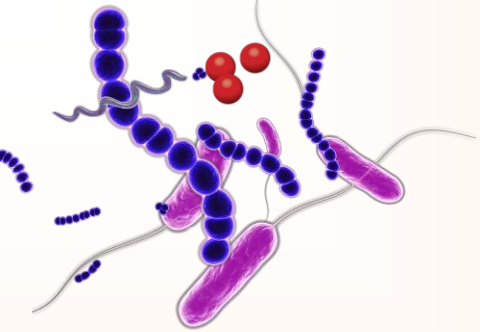
- Understand how the intestinal microbiome changes energy acquisition and storage
- Determine differences associated with efficient and inefficient animals
- Select bacterial candidates
- Inoculate animals



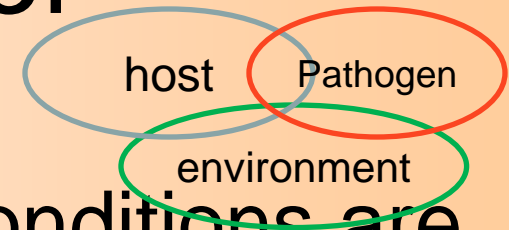
# Complex communication circuits within the intestine

(Hooper et al. BioEssays 20:336–343, 1998)

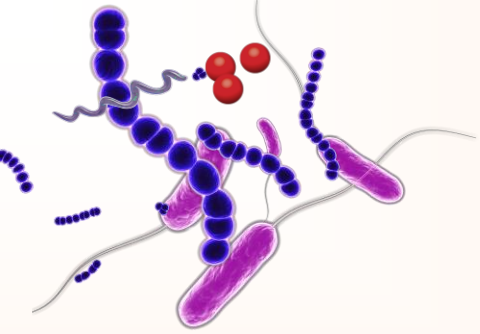




# Regulation of Pathogen Behavior



- Disease triad - symptoms of disease occurs only when conditions are conducive
  - Host condition includes the microbial community
  - Environment includes microbial community
  - Pathogen responds to conditions established by the host and to that produced by microbiome

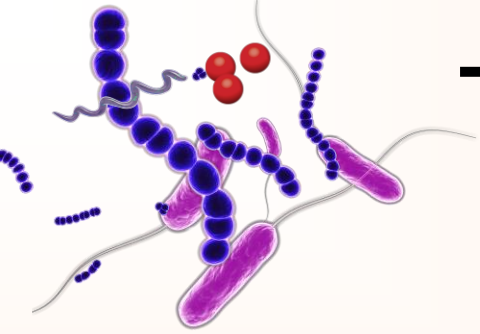


# Mechanisms for producing disease

- Invade and multiply within host
- Produce toxins



**Or Both!**

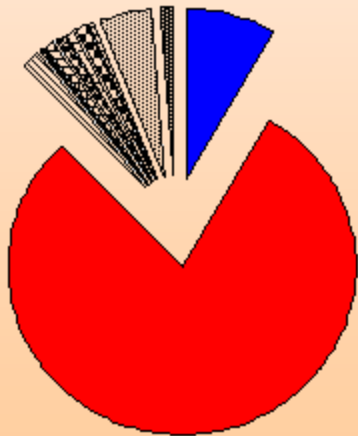


# This is especially true for the Clostridia

- Degradative organisms whose enzymes may be toxic to host tissue
- Secrete enzymes when rapidly growing
- Exhibit reduced growth rates in response to quorum sensing
- Exhibit reduced toxin production in response to quorum sensing
- Significant pathogens (necrotic enteritis, dysbacteriosis, nonspecific enteritis, gangrenous dermatitis)

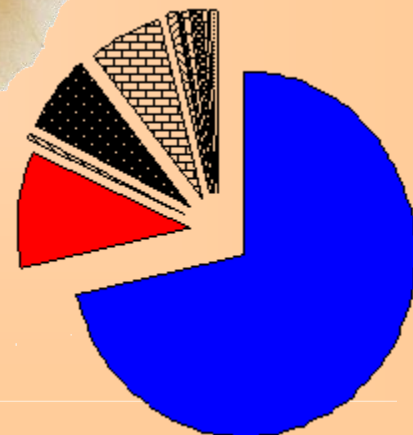
# Microbial Composition of the Broiler Intestine (16S based)

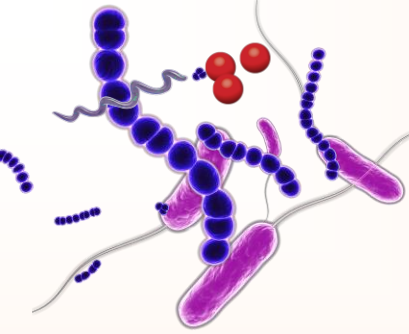
**Cecum**



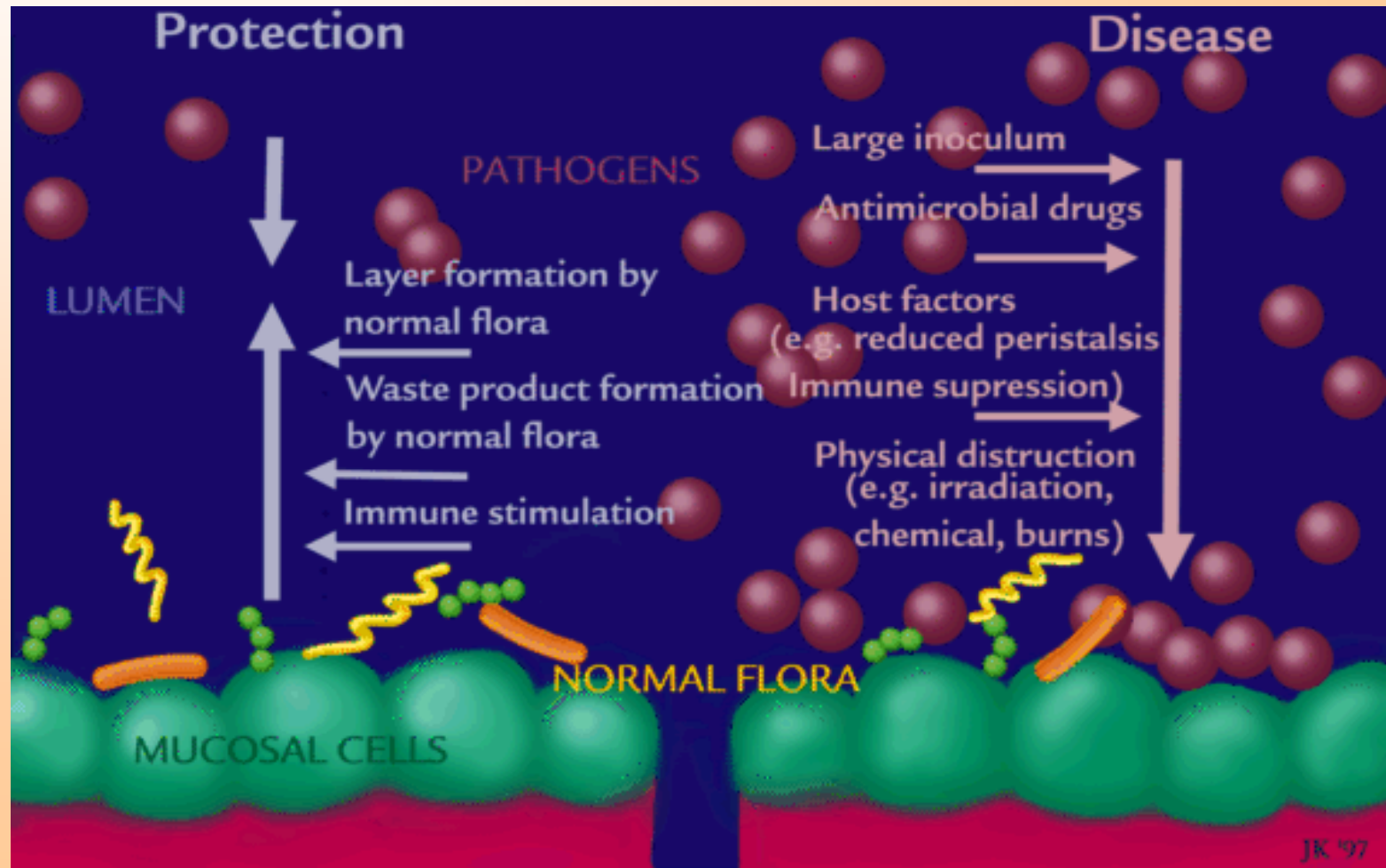
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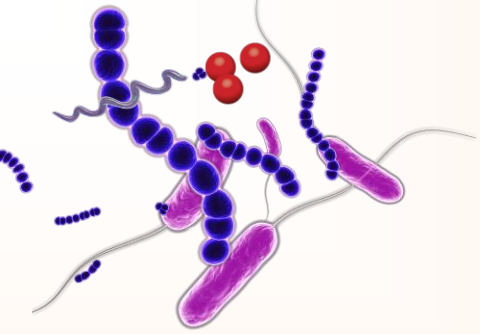
**Ileum**





Disease = a breakdown of normal host defenses.





# Mechanism of Action of Microbiome Modulators

- Bacterial growth modifiers, probiotics, feed enzymes, oligosaccharides
- possible effects on intestinal bacterial community
  - Density
  - Composition
  - Metabolism
  - Pathogenicity

